ATTACHMENT 1

Flavonoid composition of tea: Comparison of

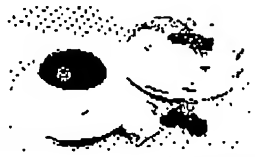
S. Bhagwat[1], G.R. Beecher[1], D.B. Haytowitz[1], J.M. Holden[1], J. Dwyer[2], J. F
A.L. Eldridge[3], S. Agarwal[4], and D.A. Balentine[4]
[1]Beltsville Human Nutrition Research Center, ARS, USDA, Beltsville, MD 20705. [2]Jean Mayer
Schools of Medicine and Nutrition, Boston, MA. [3]General Mills, M
[4]Unilever Bestfoods North America, Englewood Cliffs

Abstract

Tea, the most widely consumed beverage in the world, is produced from the leaves of Camellia sinensis. The type of tea produced depends on the length of fermentation of the leaves–green tea is not fermented, black tea is nearly completely fermented, while Oolong tea is partially fermented. The prominent flavonoids in the tea are the flavan-3-ols catechin, epicatechin, epicatechin gallate, epigallocatechin, and epigallocatechin gallate and their fermentation products, derived tannins–theaflavins and thearubigins. Some epidemiological studies, clinical trials and animal studies have shown association between tea drinking and reduced risk of chronic diseases, particularly cardiovascular diseases and some cancers. To develop a database on the flavonoid content of teas to aid researchers in assessing the dietary intake of these compounds. We have collected and evaluated analytical data on all teas through extensive literature searches and new analytical data provided by Unilever Bestfoods North America. Data analysis revealed the following: Dry green and black teas contain comparable amounts of total flavonoids, but green tea contains mostly catechins, about 3.5 times that of black tea. As tea leaves are fermented, theaflavins and thearubigins increase, while catechins decrease. Black tea has 93 times more theaflavins and 45 times more thearubigins compared to green tea. Decaffeination reduces total catechins in both black and green dry teas by about 15 times and 3 times respectively. Decaffeinated green tea contains comparable amounts of total catechins (3,942mg/100g) as in regular black tea (3,606 mg/100g). The flavonols quercetin, kaempferol and myricetin are not affected either by fermentation or by decaffeination. All ready to drink teas, black, green or made from powder have approximately a fifth to a tenth of the flavonoids compared to similar teas brewed from leaves or tea bags. The type of tea and the preparation techniques must be considered in assessing tea flavonoid contents.

Introduction

Flavonoids are biologically active polyphenolic compounds widely distributed in plants. Food sources of flavonoids are vegetables, fruits, nuts, seeds, roots, and beverages like tea and wine. Health effects of tea have been recognized since the ancient times. In a review on the role of tea in human health McKay and Blumberg (2002) noted that tea polyphenols could play a role in the prevention of cancer and heart disease. To date, the most protective effect is shown against lung cancer (Chung, 2002). The major antioxidant activity of tea extracts is attributed to Epigallocatechingallate (EGCG) (Fourneau, et al 1996). Besides the catechin molecule, which is a potent antioxidant, gallate moiety is an important contributor to the antioxidant activity (Hatbowy and Balentine 1997). Arts, et al (2001) observed an inverse relationship between tea consumption and ischemic heart disease, but not stroke, in the Zutphen elderly study. The USDA Database for the Flavonoid Content of Selected Foods, released in March 2003, contains information on the most prevalent dietary flavonoids.



Methods

Analytical data were provided in cooperation with Unilever Bestfoods, North America, Englewood Cliffs, New Jersey.
- Commercial tea and tea products were purchased in 3 regions of the United States. Two different grocery stores were selected in each city
  - East - Washington, D.C.;
  - West - Los Angeles; and
  - Midwest - Chicago).
- Tea brands and tea products were based on market-share data obtained from A.C. Nielsen, Inc.
- Samples were shipped to Lipton laboratories for analysis.
- HPLC was used to separate and quantitate catechins and their gallate esters and flavonols. This method had been approved by the working group of International Life Sciences Institute (ILSI) (Critical Reviews in Food Science and Nutrition, Ed. Clydesdale, 2001)
- Since the reference standard for thearubigins was not available, the content of thearubigins was determined by calculation. The total polyphenols were determined by Folin-Ciocalteu method and the following formula was used to calculate thearubigins

Thearubigins = (Folin-Ciocalteu Polyphenols mg - (total catechins mg * 1.150) + (total theaflavins mg * 1.500) + (kaempferol mg * 1.400) + (myricetin mg * 0.94) + (quercetin mg * 0.900) - (gallic acid mg + theogallin mg + cinnamic acid mg)) * 1.464
(Balentine, unpublished)

Literature searches were done using key words for flavonoids and tea (Camellia, thea, tea) from various databases.
- The relevant articles were reviewed and articles containing analytical data were retrieved.
- Data from analytical studies which used spectrophotometry alone, Flavognost method, paper or thin layer chromatography for separation or quantification of tea compounds were rejected for the inclusion in the database. Only one article reported data on flavone-C-glycosides and was therefore set aside.

Data Evaluation and Compilation

The NDL has developed new software for evaluating data quality (Holden et al 2002) based on the criteria described earlier (Mangels, et al 1993). Each value for each compound is evaluated for the following criteria:
- Sampling plan
- Sample handling
- Number of samples
- Analytical method
- Analytical quality control

Critical analytical steps to aid in the evaluation for each of the five categories have been developed. Data values are rated on a scale of 0-20 for each of the five criteria. Ratings are combined over all sources for a single compound to yield a Confidence Code (CC). The algorithm for combining ratings from the five categories of the data aggregation has been revised to avoid the possibility that the aggregation of several mediocre data points would together merit the higher CC rating which is the indicator of data quality.

The data were aggregated according to the Nutrient Data Bank number (NDB) for each food and the mean value (mg/100g) determined. The standard error of the mean (SEM), minimum (Min.) and maximum (Max.) values for each flavonoid compound for each food along with data quality rating are also included in the database. Values are reported on the as consumed (brewed) basis.

Table 1. Flavonoid content of black tea

Tea, black, brewed		
Subclass	Flavonoid	Mean
Flavan-3-ols	Catechin	1.20
	Epicatechin	2.34
	Epicatechin gallate	7.15
	Epigallocatechin	9.23
	Epigallocatechin gallate	10.21
	Gallocatechin	1.20
	Theaflavin	1.58
	Theaflavin-3-3-digallate	1.03
	Theaflavin-3-gallate	1.51
	Theaflavin-3-gallate	1.25
	Thearubigins	73.84
Flavonols	Kaempferol	1.29
	Myricetin	0.46
	Quercetin	2.18

Tea, black, brewed, decaffeinated		
Subclass	Flavonoid	Mean
Flavan-3-ols	Epicatechin	0.49
	Epicatechin gallate	0.64
	Epigallocatechin	0.15
	Epigallocatechin gallate	1.01
	Theaflavin	0.35
	Theaflavin-3-3-digallate	0.43
	Theaflavin-3-gallate	0.19
	Theaflavin-3-gallate	0.21
	Thearubigins	96.03
Flavonols	Kaempferol	1.25
	Myricetin	0.31
	Quercetin	2.84

Tea, black, ready-to-drink, plain and flavo		
Subclass	Flavonoid	Mean
Flavan-3-ols	Epicatechin	0.49
	Epicatechin gallate	0.91
	Epigallocatechin	0.85
	Epigallocatechin gallate	0.71
	Theaflavin	0.15
	Theaflavin-3-3-digallate	0.04
	Theaflavin-3-gallate	0.07
	Theaflavin-3-gallate	0.06
	Thearubigins	25.46
Flavonols	Kaempferol	0.86
	Myricetin	0.97
	Quercetin	0.71

Table 2. Flavonoid content of green tea

Tea, green, brewed		
Subclass	Flavonoid	Mean
Flavan-3-ols	Catechin	2.15
	Epicatechin	8.56
	Epicatechin gallate	21.94
	Epigallocatechin	15.72
	Epigallocatechin gallate	86.42
	Theaflavin	0.04
	Theaflavin-3-3-digallate	0.01
	Theaflavin-3-gallate	0.01
	Theaflavin-3-gallate	0.03
	Thearubigins	1.00
Flavonols	Kaempferol	1.47
	Myricetin	1.10
	Quercetin	2.63

tea: Comparison of black and green teas

Haytowitz[1], J.M. Holden[1], J. Dwyer[2], J. Peterson[2], S.E. Gebhardt[1],
ridge[3], S. Agarwal[4], and D.A. Balentine[4]
ISDA, Beltsville, MD 20705. [2]Jean Mayer USDA HNRC on Aging and Tufts University
I Nutrition, Boston, MA. [3]General Mills, Minneapolis, MN,
stfoods North America, Englewood Cliffs, NJ

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Table 1. Flavonoid content of black tea

Subclass	Flavonoid	Mean	SEM	N	Min	Max	CC
Tea, black, brewed							
Flavan-3-ols	Catechin	1.40	0.12	62	0.35	4.79	B
"	Epicatechin	2.34	0.18	77	0.48	8.74	B
"	Epicatechin gallate	7.15	0.43	77	2.00	18.98	B
"	Epigallocatechin	9.23	0.90	77	0.29	31.04	B
"	Epigallocatechin gallate	10.31	0.96	77	1.14	40.68	B
"	Gallocatechin	1.26	0.23	16	0.56	2.70	A
"	Theaflavin	1.58	0.16	38	0.35	5.27	B
"	Theaflavin-3,3'-digallate	1.75	0.21	39	0.08	4.08	B
"	Theaflavin-3'-gallate	1.51	0.18	39	0.12	4.13	B
"	Theaflavin-3-gallate	1.25	0.14	39	0.06	3.19	B
"	Thearubigins	74.44	7.81	31	48.28	138.60	B
Flavonols	Kaempferol	1.29	0.09	44	0.25	2.41	B
"	Myricetin	0.48	0.04	33	0.17	0.90	A
"	Quercetin	2.19	0.20	44	0.41	4.75	B
Tea, black, brewed, decaffeinated							
Subclass	Flavonoid	Mean	SEM	N	Min	Max	CC
Flavan-3-ols	Epicatechin	0.49	0.13	4	0.34	0.67	B
"	Epicatechin gallate	0.64	0.38	4	0.25	1.73	B
"	Epigallocatechin	0.55	0.18	4	0.38	1.03	B
"	Epigallocatechin gallate	1.01	0.48	4	0.49	2.45	B
"	Theaflavin	0.33	0.10	4	0.08	0.66	B
"	Theaflavin-3,3'-digallate	0.43	0.37	4	0.00	1.52	B
"	Theaflavin-3'-gallate	0.18	0.15	4	0.00	0.61	B
"	Theaflavin-3-gallate	0.41	0.24	4	0.11	1.14	B
"	Thearubigins	49.03	1.13	4	48.05	51.52	B
Flavonols	Kaempferol	1.25	0.20	4	1.00	1.84	B
"	Myricetin	0.33	0.05	4	0.26	0.49	B
"	Quercetin	2.84	0.20	4	2.40	3.30	B
Tea, black, ready-to-drink, plain and flavored							
Subclass	Flavonoid	Mean	SEM	N	Min	Max	CC
Flavan-3-ols	Epicatechin	0.40	0.15	17	0.00	2.66	A
"	Epicatechin gallate	0.27	0.09	17	0.00	0.67	A
"	Epigallocatechin	0.85	0.42	17	0.00	7.45	A
"	Epigallocatechin gallate	0.51	0.18	17	0.00	3.11	A
"	Theaflavin	0.05	0.02	17	0.00	0.18	A
"	Theaflavin-3,3'-digallate	0.04	0.02	17	0.00	0.31	A
"	Theaflavin-3'-gallate	0.02	0.01	17	0.00	0.09	A
"	Theaflavin-3-gallate	0.06	0.02	17	0.00	0.27	A
"	Thearubigins	25.49	3.47	17	7.00	56.70	A
Flavonols	Kaempferol	0.69	0.08	17	0.14	1.23	A
"	Myricetin	0.87	0.09	17	0.11	1.46	A
"	Quercetin	0.74	0.15	17	0.20	2.10	A

Table 2. Flavonoid content of green tea

Subclass	Flavonoid	Mean	SEM	N	Min	Max	CC
Tea, green, brewed							
Flavan-3-ols	Catechin	2.85	1.71	38	0.00	44.40	B
"	Epicatechin	8.86	0.83	62	1.80	26.00	B
"	Epicatechin gallate	21.96	3.09	52	1.30	76.00	B
"	Epigallocatechin	18.72	1.78	62	1.00	54.40	B
"	Epigallocatechin gallate	88.32	7.29	62	18.13	204.50	B
"	Theaflavin	0.05	0.01	4	0.02	0.08	B
"	Theaflavin-3,3'-digallate	0.01	0.01	4	0.00	0.03	B
"	Theaflavin-3'-gallate	0.01	0.01	4	0.00	0.01	B
"	Theaflavin-3-gallate	0.01	0.01	4	0.00	0.03	B
"	Thearubigins	1.08	1.08	4	0.00	4.30	B
Flavonols	Kaempferol	1.42	0.22	12	0.87	3.31	B
"	Myricetin	1.10	0.11	12	0.52	1.60	B
"	Quercetin	2.69	0.26	12	1.40	4.10	B

Table 3. Total flavonoid subclasses for teas

Tea Product	Catechins, Total	Theaflavins, Total	Thearubigins, Total	Flavonols, Total
Black, dry, leaves	3605.63	603.28	5918.94	371.27
Black, decaffeinated	242.30	123.10	4412.61	398.48
Black, brewed	34.26	6.09	73.44	3.86
Green, dry, leaves	17515.98	6.78	131.91	515.70
Green, decaffeinated	3942.16	26.70	972.52	444.65
Green, brewed	132.12	0.07	1.08	112.11
RTD* black	2.06	0.17	25.49	2.27
RTD, green	11.66	0.04	0.0	1.58
RTD made with powder	2.02	0.03	23.65	1.40

* Ready-to-drink

Results

Literature Review:

- Approximately 79 articles had descriptions about analysis of tea and flavonoids
- Approximately 25 articles had analytical quantitative data
- 14 articles had acceptable analytical data
- Additional analytical data were provided by Unilever Bestfoods

Discussion:

- Dry green tea contains mostly catechins (3.5 times that of black dry tea)
- Dry black tea contains 99 times more theaflavins and and 45 time more thearubigins compared to dry green tea.
- Quercetin is the prominent flavonol in tea.
- Decaffeination reduces catechins in both dry black (14 times) and dry green(3 times) teas considerably.
- Decaffeination or fermentation does not affect flavonol levels
- All ready to drink teas contain 5th to 10th of the flavonoids compared to their respective brewed teas.

Database:

- Table sorted by food and flavonoid class
- Confidence code and number of each aggregated flavonoid provided for each code
- Database is available on NDL's Web site http://www.nal.usda.gov/fnic/foodcomp

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